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Short communication

Toxic and essential trace metals in muscle, liver and kidney of bovines from a polluted area of Morocco

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Abstract

Toxic and essential trace metals were measured in muscle, bone, liver and kidney of bovine grazing on the municipal wastewater spreading field of Marrakech City (Morocco). Bovines were found to be seriously contaminated by toxic metals, especially cadmium, and levels were higher in liver and kidney, specific target organs for metal bioaccumulation. The high cadmium content seemed to contribute to a reduction in zinc and copper levels. The arithmetic mean concentrations of zinc, copper and cadmium in liver and kidney were respectively: 126, 112 and 5.1 mg/kg in liver; 89, 33 and 10.3 mg/kg in kidney.

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1. Introduction

General concern about the impact on human health of exposure to environmental pollution has led to increasing attention over the last two decades to the presence of toxic substances in the human diet. To our knowledge, none of these investigations has concerned Morocco.

In the region of Marrakech City investigated in this study, livestock feed mainly on wastewater and wastewater-irrigated lucerne and corn leaves. The benefits of using wastewater as a fertiliser have been well-documented. However, a number

of ecotoxicological concerns arise from such a practice. The risk to humans through consumption of vegetable crops and animal products is apparent (Sedki et al., 1995a, 1995b). One of the more pressing concerns is that of trace elements (Lekouch et al., 1999, 2001).

Several investigations have studied the transfer of heavy metals from soil to animals either by direct contamination or via the vegetation (Penumarthi and Oehme, 1980; Medeiros et al., 1988). Even though soil is a biochemical reactor acting as an active filter for trace metals (Hogue et al., 1984), these may accumulate in the vegetation and, consequently, create a risk for animals and humans. Previous ecotoxicological studies on pigs,

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sheep and cattle have shown a direct correlation between metal concentrations found in animal feed and in animal tissues (Medeiros et al., 1988; Baxter et al., 1982).

The main aim of the present study was to investigate the presence of trace metals in muscle, bone, liver and kidney of bovine reared and slaughtered in the wastewater spreading field of Marrakech City (Morocco) from 1998 to 2000. Trace elements were measured and levels compared with those reported in other countries. Interactions between toxic (cadmium) and essential (zinc and copper) trace elements were discussed.

2. Materials and methods

The Marrakech City wastewater spreading field consists of a triangular area of 3000 ha of which 2000 ha are irrigated with untreated wastewater. This water is polluted with heavy metals such as cadmium, lead and chromium (Sedki and Pihan, 1991; Sedki et al., 1995b) originating mainly from industry. Local industries include craft and food industries. The latter do not contribute to increased levels of heavy metals, whereas craft industries using chemical products (fungicides, bactericides...) in the treatment of wool and leather, for instance, are probably responsible for the high cadmium and chromium content of wastewater. Corrosion of the drinking water distribution system in the old city is also responsible for the release of lead (Pihan et al., 1987).

Each year of the study (1998, 1999 and 2000), 10 cattle reared in the spreading field were analysed. Samples were taken from liver, kidney, femur muscle and femur bone. These were oven-dried (80 °C) to a constant weight and then powdered. Accurately weighed aliquots of approximately 100 mg of each powdered organ were digested with 2 ml of concentrated nitric acid (HNO₃) at 250 °C for 4 h. The volume was adjusted to 20 ml with double distilled water.

2.1. Analytical method

Trace elements were determined by graphite furnace atomic absorption spectrophotometry (GFAAS) using a Perkin Elmer Spectrometer Model

5000 instrument equipped with deuterium arc background correction. Each sample was analysed in duplicate. Control samples were provided from a non-polluted rural zone located 20 km from the spreading field. Levels were expressed as micrograms per gram.

Calibration standards were prepared each day using a manual standard addition procedure.

All metal determinations were carried out in the Toxicology Laboratory of the University of Nantes from France.

2.2. Limit of detection

The limit of detection calculated following the recommendations of the International Atomic Energy Agency (IAEA, 1995) was 0.06 µg/g for cadmium.

2.3. Data analysis

Data were analysed statistically by the arithmetic mean, standard deviation, Pearson linear correlation, Student test, and Newman and Keuls test. The level of significance was set at 5%.

3. Results

Table 1 shows the results (arithmetic mean, standard deviation and range) obtained for the essential and toxic trace metals analysed. Table 2 compares these values with previous studies on bovines in other countries (The Newman and Keuls test for 5%).

The zinc levels were 89 ± 13.6 µg/g in kidney and 129 ± 19 µg/g in bone. The difference between zinc levels in kidney and other organs was significant. The highest mean copper level (112 ± 22.9 µg/g) was observed in liver. The difference with other organs was significant ($P < 0.05$). In muscle, kidney and bone, mean copper levels were 4.4 ± 1.1 µg/g, 32.7 ± 7 µg/g and 2.3 ± 0.6 µg/g, respectively.

A high level of cadmium was found in kidney (10.1 ± 2.5 µg/g), whereas mean liver cadmium level was 5.2 ± 1.4 µg/g. Statistical analysis showed a significant difference ($P < 0.05$) between cadmium levels in kidney and other organs.

Table 1
Mean levels of trace elements in tissues of cattle (mg/kg dryweight)

		Kidney		Liver		Bone		Muscle	
		Cattle	Control	Cattle	Control	Cattle	Control	Cattle	Control
Zn	M	89	39.8	126	59.7	129	75.7	123	45.8
	SD	13.6	3.6	22.2	7.1	19.3	5.4	21.4	5.9
	CV	15.2	9	17.6	11.9	14.9	7.1	17.3	12.8
	Mn	66.0	36.2	95.0	52.6	96.0	70.3	89.0	39.9
	Mx	112	43.4	161	66.9	155	81.1	154	51.7
Cu	M	33.2	15.3	112	45	23.3	16.2	4.4	1.2
	SD	7	1.2	22.9	5.5	6.3	2.6	1.1	1.0
	CV	21	7.8	20.4	12.2	27	16	25	10
	Mn	28	14.1	76	39.5	16.9	13.5	2.6	1.0
	Mx	48	16.5	156	50.5	36.2	18.8	6.0	1.3
Cd	M	10.3	2.2	5.1	1.7	0.47	0.2	0.6	0.2
	SD	2.5	0.8	1.4	1.1	0.02	0.02	0.2	0.07
	CV	24.2	38.6	27.4	64	5.2	10	33.5	35
	Mn	5.7	1.4	2.9	0.6	0.45	0.2	0.25	0.15
	Mx	13.5	3.1	7.1	2.7	0.5	0.25	1.0	0.3

M: mean; Mn: minimum; S.D.: standard deviation; Mx: maximum; CV: coefficient of variation; Control: control cattle.

Table 2
Comparison of zinc in tissues of bovine from Morocco and other countries (mg/kg dryweight)

Years Metal	Muscle			Liver			Kidney			References
	Zn	Cu	Cd	Zn	Cu	Cd	Zn	Cu	Cd	
1984	25	0.7	0.12	33	17	0.17	17	4.3	4.3	Falandysz, 1993
1987	33	1.5	0.06	38	26	0.12	22	5.4	0.5	Falandysz, 1993
1988	35	0.8	—	50	33	0.15	23	5.8	0.5	Falandysz, 1993
1986	—	—	—	115	118	0.11	119	17	0.54	Friel, 1987
1981	250	2.9	0.12	141	113	21.6	131	76	6	Baxter et al., 1982
1982	247	3.9	0.01	113	114	0.8	76	113	5.9	Baxter et al., 1983
2000	123	4.4	0.1	126	112	5.1	89	33	10	This study

In comparison to control animals, experimental bovine had significantly higher levels of metals (Table 1). The pollution factor (PF), defined as element concentration in experimental animals by element level in control animals was always great-

er than 1 (Table 4). Values were particularly high for cadmium.

Higher coefficients of variation (CV) were observed for cadmium in all the organs analysed

Table 3
Heavy metal content in wastewater, soil and plants

	Wastewater (mg/l)	Soil (mg/g)	Lucerne (mg/g)	Corn leaves (mg/g)
Zn	4.5 " 1.5	450 " 7.5	250 " 4.5	380 " 6.6
Cu	0.4 " 0.2	120 " 22	44 " 12	65 " 14
Cd	0.03 " 0.01	5.5 " 2.5	1.4 " 1.1	2.2 " 1.3

Table 4
Comparison of metal concentrations in animal tissues and plants (concentration factor-CF) and in control bovine and experimental bovine (pollution factor-PF)

CF/yPF	Kidney	Liver	Bone	Muscle
Zn	0.3y2.2	0.5y2.1	0.5y1.7	0.4y2.6
Cu	0.8y2.1	2.8y2.4	0.05y1.4	0.1y3.6
Cd	3.3y4.6	0.9y3.0	0.1y2.3	0.1y3.0

Table 5

Relationship between concentrations of cadmium and essential metals in cattle organs

Organs	Zn/Cd	Cu/Cd
Kidney	8.64	3.2
Kidney*	18.01	6.9
Liver	24.7	21.9
Liver*	35.1	26.5
Muscle	205	7.3
Muscle*	229	6
Bone	274.4	49.3
Bone*	378.5	81

* Control bovine.

in both cattle and sheep reflecting the heterogeneity of individual levels of cadmium.

Trace element levels were also measured in lucerne and corn leaves grown in the area of study and used in rearing. The results (Table 3) show the presence of metals in the plants due to the accumulation of these elements in the soil. A concentration factor (CF), defined as the concentration of metals in animal tissues/the concentration in plants, was calculated to measure the transfer of pollutants through the food chain. As shown in Table 4, concentration factors for cadmium in kidney and for copper in liver were greater than 1. The values for cadmium were especially high.

4. Discussion

As is the case for many trace elements, metal levels in tissue are largely dependent on the metal content of diet. The relationship between orally administered cadmium and tissue content was estimated by Vos et al. (1988) as 0.9% based on an experiment in which pregnant ewes and lambs were fed a diet containing from 0.2 to 0.4 mg/kg cadmium. This estimate is considerably lower than the 9% apparent absorption calculated by Doyle et al. (1974) for lambs receiving a diet containing 60 mg Cd/kg (Table 5).

The results of this study show that bovines grazing on the municipal wastewater spreading field of Mirrakech are concentrating the trace elements investigated. The copper levels observed in the liver are similar to those reported by Baxter

et al. (1982) (113 mg/kg) and by Medeiros et al. (1988) (119 mg/kg) in cattle reared on sewage sludge. The mean levels of zinc observed in the liver are lower than those reported by Frielet al., 1987 (161 mg/kg) and by Baxter and Kienholz (1983) (212 mg/kg) for cattle. However, these values are greater than those (50 mg/kg) reported by Underwood (1977) in liver of cattle living in spreading fields.

The cadmium level in kidney tissues was significantly higher in experimental animals than controls. The mean levels observed were lower than those found by Baxter and Kienholz (1983) in cattle (55 mg/kg) and sheep (16 mg/kg) exposed to environmental contamination. Baxter and Kienholz (1983) reported levels reaching 20 mg/kg in kidneys of bovine exposed to sewage sludge. Van-Der-Veen and Vroman (1986) reported cadmium levels of 12 mg/kg in lambs exposed to contaminated feed. These results are higher than those (2 mg/kg) noted by Frielet al. (1987) in cattle kidney from animals feeding on a non-polluted soil.

The results show cadmium to concentrate primarily in the kidney, Cu in the liver and Zn in the kidney, liver and muscle. This unequal distribution amongst the organs is related to differences in the specific physiological functions of these elements and depend on their relative abundance in intracellular ligandable to bind metals, such as metalloproteins (Koh and Judson, 1986; Kagi and Kojima, 1987).

The Zn/Cd and Cu/Cd ratios observed in kidney, liver, bone and meat were higher in control animals indicating that cadmium represses these essential metals (Table 5). As could be expected from the role of kidney in alleviating cadmium accumulation, the Zn/Cd ratio in this organ was much lower than in liver, bone and muscle. Most authors found that cadmium caused the greatest degree of disruption in copper and zinc homeostasis. A significant positive association between renal cadmium and zinc residues and a significant negative correlation between kidney cadmium and copper levels have also been reported (Alonso et al., 2002).

The consumption of meat, liver and kidney from livestock reared in this polluted area certainly contributes significantly to the average daily metal

intake by man. According to the World Health Organisation, the weekly cadmium intake by man should not exceed 400–500 mg (WHO, 1980). In Morocco, no regulations have yet been established for heavy metals in meat or bovine organs but this issue has become important in many countries.

5. Conclusions

The concentrations of cadmium, copper and zinc found in muscle, liver, kidneys and bone of live-stock grazing on the municipal sewage spreading field of Mirrakesh are very high. High levels of cadmium were encountered resulting from bio-accumulation (CF) and bio-amplification (PF) processes. The highest levels of metals were found in liver and kidney, which are known specific target organs of metal bioaccumulation. This study showed that the high cadmium levels measured in the organs of cattle and sheep seemed to disrupt normal trace element metabolism, decreasing zinc and copper levels.

As cadmium is known to be a highly toxic compound to which chronic exposure results in severe diseases or even death, there is an urgent need to initiate an extensive epidemiological study of people consuming products (vegetables and meat) originating from this area.

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